

COVER ESTIMATES OF EPIPHYTIC CORALLINE ALGAE (CORALLINALES, RHODOPHYTA): BRAUN-BLANQUET VS COMPUTER IMAGE ANALYSIS

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ABSTRACT — Two techniques for examining percentage cover of epiphytic nongeniculate coralline algae were investigated. Percentage cover of *Pneophyllum fragile* Kützinger, *Melobesia membranacea* (Esper) Lamouroux and *Melobesia rosanoffii* (Foslie) Lemoine growing on *Laurencia elata* (C. Agardh) Hooker & Harvey (Rhodomelaceae, Ceramiales), was estimated using a modified Braun-Blanquet rating scheme and with computer image analysis (digitised cover method: DCM). Using an artificial paper pattern DCM was found to be accurate in assessing known "percentage cover". Cover of epiphytic nongeniculate corallines was more accurately determined by DCM than by the Braun-Blanquet rating system. The Braun-Blanquet method consistently yielded over-estimates when cover was high, and under-estimates when it was low. Using DCM rather than the Braun-Blanquet method, percentage cover is measured, not estimated. DCM may have broader applications for both non-algal epiphytes and non-epiphytic systems.

RÉSUMÉ — Deux techniques de détermination du pourcentage de couverture de corallines non articulées épiphytes ont été comparées. Les pourcentages de couverture de *Pneophyllum fragile* Kützinger, *Melobesia membranacea* (Esper) Lamouroux et *Melobesia rosanoffii* (Foslie) Lemoine épiphytes de *Laurencia elata* (C. Agardh) Hooker & Harvey (Rhodomelaceae, Ceramiales), ont été estimés, à l'aide d'un système de codage de Braun-Blanquet modifié, d'une part, et à l'aide d'un système d'analyse d'image par ordinateur (*Digitised Cover Method*: DCM), d'autre part. En utilisant une configuration artificielle en papier, le système DCM s'est révélé performant puisqu'il a déterminé les "pourcentages de couvertures" connus. La couverture des corallines non articulées épiphytes a été déterminée avec plus de précision par la technique du DCM qu'avec l'aide du système de Braun-Blanquet. La méthode de Braun-Blanquet a mené systématiquement à des surestimations lorsque le pourcentage de couverture était élevé et à des sous-estimations lorsque ce pourcentage était faible. Lorsque l'on utilise la méthode du DCM, le pourcentage de couverture est mesuré et non estimé. La méthode du DCM pourrait avoir des applications plus larges pour des épiphytes autres que des algues et pour des organismes non épiphytes.

KEY-WORDS: Braun-Blanquet, digitised cover method (DCM), epiphyte, image analysis, *Laurencia*, nongeniculate coralline algae, percentage cover, *Pneophyllum*, *Melobesia*.

INTRODUCTION

Nongeniculate coralline algae (Corallinaceae, Rhodophyta) commonly occur as epiphytes on various marine plants worldwide (Steneck, 1986; Woelkerling, 1988; Kjøstørud, 1997). Studies of their ecology generally involve analysis of changes or patterns in their percentage cover of the substrate. So far, cover has been estimated using various modifications of the subjective rating system developed by Braun-Blanquet (1928) (e.g. Ballantine, 1979; Jacobs *et al.*, 1983; Heijs, 1985; Kendrick *et al.*, 1988; Otero-Schmitt & Pérez-Cirera, 1996). This method has advantages of speed (Kershaw, 1973; Kendrick *et al.*, 1988), but it is a scale which lacks measurement (Mueller-Dombois & Ellenberg, 1974). Furthermore, depending on the level of cover, inaccuracy and unconscious bias may limit its usefulness (Kershaw, 1973; Greig-Smith, 1983).

Recently Morcom *et al.* (1997) used image analysis to measure percentage cover of nongeniculate corallines on *Laurencia elata*. This, however, has raised unanswered questions such as: does image analysis have advantages and/or limitations when compared with Braun-Blanquet schemes, and is image analysis more accurate than Braun-Blanquet estimates?

In this study, Braun-Blanquet and computer image analysis (the digitised cover method: DCM) are assessed, first by using artificial paper patterns to determine the accuracy of the digitised cover method, and second, by comparing Braun-Blanquet and DCM estimates of cover in a natural system — nongeniculate corallines epiphytic on the red alga *Laurencia elata* (C. Agardh) J.D. Hooker & Harvey (Rhodomelaceae, Ceramiales).

MATERIALS AND METHODS

To determine the accuracy of the digitised cover method (DCM) we compared digitised estimates of percentage cover with known percentage cover values (0.9%, 4.5%, 14%, 17%, 32% and 100%). Known percentage cover values were obtained using an artificial paper pattern. This was produced by cutting irregular shapes from paper, weighing them and calculating their percentage cover, then photocopying the pieces in a haphazard arrangement.

Two methods were used to assess the cover of epiphytic corallines. A modified Braun-Blanquet scheme was applied as follows. Percentage cover was estimated as: +, < 1%; 1 — $\geq 1\%$ cover < 5%; 2 — $\geq 5\%$ cover < 10%; 3 — $\geq 10\%$ cover < 20%; 4 — $\geq 20\%$ cover < 40%; 5 — $\geq 40\%$ cover < 60%; 6 — $\geq 60\%$ cover < 80%; 7 — $\geq 80\%$ cover = 100%. Three experienced experimental volunteers and the first author performed the Braun-Blanquet assessments. DCM measurements were taken by the first author.

Percentage cover was digitised using the computer image analysis package Trace (Leading Edge Pty Ltd ©). DCM involves microscopic examination with computer image analysis. When substratum (or artificial paper pattern) is viewed through a binocular dissecting microscope (16x mag.) a red dot is visible through one eyepiece. This dot is the "marker point" of a mouse attached to a digitising tablet (Summa Sketch II ®, Summa-

graphics[®]). The red dot is used to trace the edges of plaques of corallines (or simulated epiphytic cover), and the results are displayed on an adjacent monitor. Percentage cover was measured by tracing the whole area being examined (i.e. paper area or 1 cm length of *L. elata*) and recording its value, then tracing the areas covered by each coralline species (or simulated cover) within this 1 cm length or paper area and dividing by the total area.

Laurencia elata (C. Agardh) J.D. Hooker & Harvey (Rhodomelaceae, Ceramiales) is a red alga found along southern Australian rough-water coasts (for further details see Morcom *et al.*, 1997). Axes of *Laurencia elata* were randomly selected for analysis from material from one locality during February 1994: Number 16 Reef (38°25'12"S, 144°49'00"E) Rye, Victoria. This data set is independent of that analysed in Morcom *et al.* (1997).

A total of eighty 1 cm lengths of *L. elata* were examined by three experimental volunteers plus the first author. Both sides of two fronds were examined (two replicates). Each person examined twenty 1 cm lengths, and each 1 cm length was examined by two people.

Three species of nongeniculate coralline algae were found on *Laurencia elata* fronds: *Pneophyllum fragile* Kützinger, *Melobesia membranacea* (Esper) Lamouroux and *M. rosanoffii* (Foslie) Lemoine. Within the genus *Melobesia*, most plants were *M. membranacea*, but some were *M. rosanoffii*. Microscopic sectioning is required to distinguish between the two species, so results from the two were pooled; in the remainder of this paper they are referred to as *Melobesia*.

Species identification follows Saito & Womersley (1974) for *Laurencia elata* and Penrose (1996) and Woelkerling (1996) for the nongeniculate corallines. Permanent slide collections and voucher specimens (LTB 17293) are housed at LTB (Department of Botany, La Trobe University, Bundoora, Victoria, Australia), and will eventually be transferred to MEL (National Herbarium of Victoria, South Yarra, Victoria, Australia).

RESULTS AND DISCUSSION

The digitised cover method (DCM) accurately assessed known percentage cover ($r^2 = 0.997$, Fig. 1). Percentage cover estimates using DCM show a very close fit to the known cover values.

DCM showed that nongeniculate coralline algae covered 61% (*Pneophyllum* 49% and *Melobesia* 12%) of the total surface of *Laurencia elata*, non-coralline epiphytes covered 3% and the remaining 36% was unoccupied. *Pneophyllum* and *Melobesia* constitute the total coralline cover of *Laurencia elata*, where *Pneophyllum* dominates and *Melobesia* occurs sporadically. Cover within sampling units varied from absent to highly abundant, in both *Pneophyllum* (0–94%) and *Melobesia* (0–50%).

Percentage cover estimates of *Pneophyllum* and *Melobesia* (Figs 2, 3) varied between the two techniques. Comparative percentage cover estimates of *Pneophyllum* (Fig. 2) showed that below 60% cover, 95% of Braun-Blanquet estimates were lower than the DCM estimates for the same section of substrate; for estimates above 60% cover, all Braun-Blanquet estimates exceeded digitised cover estimates. Despite its infrequent occurrence, the percentage cover estimates of *Melobesia* (Fig. 3) were similar to those of *Pneophyllum*: most Braun-Blanquet estimates of percentage cover below 60% were lower than digitised cover estimates. These results highlight one of the failings of subjective rating systems, that of over- and under-estimating percentage cover (Greig-Smith,

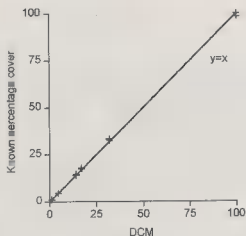


Fig. 1. Scatter plot of known percentage cover and DCM measured percentage cover. (DCM = the digitised cover method, $n = 48$; weighed paper).

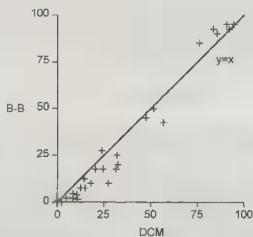


Fig. 2. Scatter plot of mean percentage cover of *Pneophyllum* (DCM = digitised cover method. B-B = Braun-Blanquet method; $n = 40$).

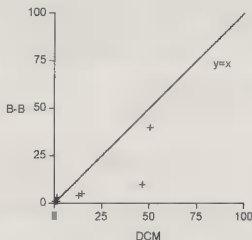


Fig. 3. Scatter plot of mean percentage cover of *Melobesia* spp. (DCM = digitised cover method, B-B = Braun-Blanquet method; $n = 40$).

Table 1. The means and variances of the proportions cover of *Pneophyllum* and *Melobesia* estimated using DCM and B-B, and their ratios (DCM = digitised cover method, B-B = Braun-Blanquet, $n = 40$).

Species	Mean			Variance		
	DCM	B-B	B-B/DCM	DCM	B-B	B-B/DCM
<i>Pneophyllum</i>	0.348	0.314	0.902	0.098	0.120	1.224
<i>Melobesia</i>	0.143	0.093	0.650	0.040	0.020	0.500

1983). Moreover when "species vary in conspicuousness, it is difficult to avoid overrating conspicuous species and underrating inconspicuous ones" (Greig-Smith 1983, p. 3).

Table 1 shows that overall the Braun-Blanquet method underestimated cover by 9.8% for *Pneophyllum* and 35% for *Melobesia*. It also underestimated the variance by 50%. This result (cover), particularly for *Melobesia*, may reflect small plaque size coupled with a sporadic distribution pattern and the associated difficulty of conspicuous species and inconspicuous ones (see Fig. 3).

When measuring cover of nongeniculate corallines, the digitised cover method was more accurate than the Braun-Blanquet method and thus eliminated much of the associated subjective error. However, there are limitations associated with DCM, including the time taken, and the "suitability" of the epiphyte and substratum. Using DCM was time-consuming (1 cm/5 mins), but improved accuracy and the ability to measure rather than to estimate percentage cover may remove "time" as a limitation. When using

DCM, some variability was observed when measuring cover repeatedly on lengths. Potential difficulties associated with cylindrical substrates have yet to be assessed.

By using DCM rather than the Braun-Blanquet approach, percentage cover is measured and not estimated. Conclusions drawn from the Braun-Blanquet data should be considered with caution (see Table 1, means & variances). Analysis using DCM appears useful for encrusting epiphytes on seagrasses or on algae with a more or less flat surface.

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